09/882,509

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     FILE 'MEDLINE, EMBASE, BIOSIS, BIOTECHDS, SCISEARCH, HCAPLUS, NTIS,
     LIFESCI' ENTERED AT 11:37:16 ON 10 DEC 2004
L1         41164 S STREPTOKINASE?
L2           364 S "S. EQUISIMILIS"
L3           103 S L1 AND L2
L4       6828585 S CLON? OR EXPRESS? OR RECOMBINANT
L5            74 S L3 AND L4
L6            29 DUP REM L5 (45 DUPLICATES REMOVED)
                 E KUPPUSAMY M/AU
L7            40 S E3
                 E LAHRI S/AU
L8             5 S E3
                 E KHATRI G S/AU
L9            55 S E3-E7
L10          100 S L7 OR L8 OR L9
L11            0 S L5 AND L10
L12            0 S L1 AND L10
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                   search transcripts to be affected by CERAB, COMPUAB, ELCOM,
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 NEWS  6  NOV 30   PHAR reloaded with additional data
 NEWS  7  DEC 01   LISA now available on STN
 NEWS  8  DEC 09   12 databases to be removed from STN on December 31, 2004

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FILE 'LIFESCI' ENTERED AT 11:37:16 ON 10 DEC 2004
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=> s streptokinase?
L1 41164 STREPTOKINASE?

=> s "s. equisimilis"
L2 364 "S. EQUISIMILIS"

=> s l1 and l2
L3 103 L1 AND L2

=> s clon? or express? or recombinant
4 FILES SEARCHED...
L4 6828585 CLON? OR EXPRESS? OR RECOMBINANT

=> s l3 and l4
L5 74 L3 AND L4

=> dup rem l5
PROCESSING COMPLETED FOR L5
L6 29 DUP REM L5 (45 DUPLICATES REMOVED)

=> d 1-29 ibib ab

L6 ANSWER 1 OF 29 EMBASE COPYRIGHT 2004 ELSEVIER INC. ALL RIGHTS RESERVED.
on STN DUPLICATE 1
ACCESSION NUMBER: 2003404343 EMBASE
TITLE: **Expression** of streptodornase by use of **streptokinase** promoter in Streptococcus equismilis H46A.
AUTHOR: Sohn H.-J.; Chin J.; Kim I.-C.; Bai S.; Lee H.B.
CORPORATE SOURCE: H.B. Lee, Department of Biological Sciences, Chonnam National University, Gwangju 500-757, Korea, Republic of. blaise@chonnam.chonnam.ac.kr
SOURCE: Korean Journal of Microbiology and Biotechnology, (2003) 31/3 (307-310).
Refs: 18
ISSN: 1598-642X CODEN: HMHAAS
COUNTRY: Korea, Republic of
DOCUMENT TYPE: Journal; Article
FILE SEGMENT: 004 Microbiology
LANGUAGE: Korean
SUMMARY LANGUAGE: English
AB A gene encoding streptodornase(sdc) from Streptococcus equisimilis H46A was **expressed** in **S. equisimilis** H46A sdc(-) under the control of the **streptokinase** gene promoter. Secretion of the streptodornase was directed by the signal sequences of

streptokinase or streptodornase. The **expressed** streptodornase activity from **S. equisimilis** H46A sdc(-) transformant with **streptokinase** promoter - streptodornase coding sequence fusion vector was 2.3 fold higher than that from wild type. Construct of signal sequence region replaced by **streptokinase** ones was similarly **expressed** as a wild type. But constructs of skc or lrp core regions of **streptokinase** promoter streptodornase fusion were similarly **expressed** as in sdc(-) mutant. In conclusion, improved **expression** of streptodornase by use of **streptokinase** promoter required the full length of promoter.

L6 ANSWER 2 OF 29 MEDLINE on STN DUPLICATE 2
ACCESSION NUMBER: 2000038313 MEDLINE
DOCUMENT NUMBER: PubMed ID: 10569766
TITLE: **Cloning**, **expression**, sequence analysis, and characterization of **streptokinases** secreted by porcine and equine isolates of Streptococcus equisimilis.
AUTHOR: Caballero A R; Lottenberg R; Johnston K H
CORPORATE SOURCE: Department of Microbiology, Immunology and Parasitology, Louisiana State University Medical Center, New Orleans, Louisiana 70112, USA.
CONTRACT NUMBER: R01DK45014 (NIDDK)
SOURCE: Infection and immunity, (1999 Dec) 67 (12) 6478-86. Journal code: 0246127. ISSN: 0019-9567.
PUB. COUNTRY: United States
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
OTHER SOURCE: GENBANK-AF104300; GENBANK-AF104301
ENTRY MONTH: 199912
ENTRY DATE: Entered STN: 20000113
Last Updated on STN: 20000113
Entered Medline: 19991220

AB **Streptokinases** secreted by nonhuman isolates of group C streptococci (Streptococcus equi, **S. equisimilis**, and S. zooepidemicus) have been shown to bind to different mammalian plasminogens but exhibit preferential plasminogen activity. The **streptokinase** genes from **S. equisimilis** strains which activated either equine or porcine plasminogen were **cloned**, sequenced, and **expressed** in Escherichia coli. The **streptokinase** secreted by the equine isolate had little similarity to any known **streptokinases** secreted by either human or porcine isolates. The **streptokinase** secreted by the porcine isolate had limited structural and functional similarities to **streptokinases** secreted by human isolates. Plasminogen activation studies with immobilized (His)(6)-tagged **recombinant** **streptokinases** indicated that these **recombinant** **streptokinases** interacted with plasminogen in a manner similar to that observed when **streptokinase** and plasminogen interact in the fluid phase. Analysis of the cleavage products of the **streptokinase**-plasminogen interaction indicated that human, equine, and porcine plasminogens were all cleaved at the same highly conserved site. The site at which **streptokinase** was cleaved to form altered **streptokinase** (Sk*) was also determined. This study confirmed not only the presence of **streptokinases** in nonhuman **S. equisimilis** isolates but also that these proteins belong to a family of plasminogen activators more diverse than previously thought.

L6 ANSWER 3 OF 29 BIOSIS COPYRIGHT (c) 2004 The Thomson Corporation. on STN DUPLICATE 3
ACCESSION NUMBER: 2000:96119 BIOSIS

DOCUMENT NUMBER: PREV200000096119
TITLE: Two **streptokinase** genes are **expressed** with different solubility in Escherichia coli W3110.
AUTHOR(S): Pupo, Elder [Reprint author]; Baghbaderani, Behnam A.; Lugo, Victoria; Fernandez, Julio; Paez, Rolando; Torrens, Isis
CORPORATE SOURCE: Biopharmaceutical Development Division, Center for Genetic Engineering and Biotechnology, Havana, Cuba
SOURCE: Biotechnology Letters, (Dec., 1999) Vol. 21, No. 12, pp. 1119-1123. print.
CODEN: BILED3. ISSN: 0141-5492.
DOCUMENT TYPE: Article
LANGUAGE: English
ENTRY DATE: Entered STN: 15 Mar 2000
Last Updated on STN: 3 Jan 2002

AB The **streptokinase** (SK) gene from **S. equisimilis** H46A (ATCC 12449) was **cloned** in E. coli W3110 under the control of the tryptophan promoter. The **recombinant** SK, which represented 15% of total cell protein content, was found in the soluble fraction of disrupted cells. The solubility of this SK notably differed from that of the product of the SK gene from **S. equisimilis** (ATCC 9542) which had been **cloned** in E. coli W3110 by using similar **expression** vector and cell growth conditions, and occurred in the form of inclusion bodies.

L6 ANSWER 4 OF 29 MEDLINE on STN DUPLICATE 4
ACCESSION NUMBER: 1999150235 MEDLINE
DOCUMENT NUMBER: PubMed ID: 10024545
TITLE: Purification and **cloning** of a **streptokinase** from Streptococcus uberis.
AUTHOR: Johnsen L B; Poulsen K; Kilian M; Petersen T E
CORPORATE SOURCE: Protein Chemistry Laboratory, Department of Molecular and Structural Biology, University of Aarhus, DK-8000 Aarhus C, Denmark.
SOURCE: Infection and immunity, (1999 Mar) 67 (3) 1072-8.
Journal code: 0246127. ISSN: 0019-9567.
PUB. COUNTRY: United States
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
OTHER SOURCE: GENBANK-AJ131604; GENBANK-AJ131605; GENBANK-AJ131631
ENTRY MONTH: 199903
ENTRY DATE: Entered STN: 19990326
Last Updated on STN: 19990326
Entered Medline: 19990312

AB A bovine plasminogen activator was purified from the culture supernatant of the bovine pathogen Streptococcus uberis NCTC 3858. After the final reverse-phase high-performance liquid chromatography step a single protein with a molecular mass of 32 kDa was detected in the active fraction. A partial peptide map was established, and degenerate primers were designed and used for amplification of fragments of the gene encoding the activator. Inverse PCR was subsequently used for obtaining the full-length gene. The S. uberis plasminogen activator gene (skc) encodes a protein consisting of 286 amino acids including a signal peptide of 25 amino acids. In an amino acid sequence comparison the **cloned** activator showed an identity of approximately 26% to the **streptokinases** isolated from Streptococcus equisimilis and Streptococcus pyogenes. Interestingly, the activator from S. uberis was found to lack the C-terminal domain possessed by the **streptokinase** from **S. equisimilis**. This is apparently a general feature of the **streptokinases** of this species; biochemical and genetic analysis of 10 additional strains of S. uberis revealed that 9 of these were highly similar to strain NCTC 3858. Sequencing of the skc gene

from three of these strains indicated that the amino acid sequence of the protein is highly conserved within the species.

L6 ANSWER 5 OF 29 BIOTECHDS COPYRIGHT 2004 THE THOMSON CORP. on STN
ACCESSION NUMBER: 1999-03949 BIOTECHDS
TITLE: Purification and **cloning** of a **streptokinase** from Streptococcus uberis; cattle plasminogen-activator purification and characterization
AUTHOR: Johnson L B; Poulsen K; Kilian M; *Petersen T E
CORPORATE SOURCE: Univ.Aarhus
LOCATION: Protein Chemistry Laboratory, Gustav Wieds Vej 10C, DK-8000 Aarhus C, Denmark.
Email: tep@mbio.aau.dk
SOURCE: Infect.Immun.; (1999) 67, 3, 1072-78
CODEN: INFIBR
ISSN: 0019-9567
DOCUMENT TYPE: Journal
LANGUAGE: English
AB A cattle plasminogen-activator was purified from the culture supernatant of Streptococcus uberis NTCTC 3858. After the final reverse-phase HPLC step, a single protein with a mol.weight of 32,000 was detected in the active fraction. A partial peptide map was established, and degenerate DNA primers were designed and used for amplification of fragments of the gene encoding the activator. Inverse polymerase chain reaction was used for obtaining the full-length gene. The S. uberis plasminogen-activator gene (skc) encodes a protein consisting of 286 amino acids including a signal peptide of 25 amino acids. In a protein sequence comparison, the **cloned** activator showed an identity of approximately 26% to the **streptokinases** isolated from Streptococcus equisimilis and Streptococcus pyogenes. The activator from S. uberis lacked the C-terminal domain possessed by the **streptokinase** from **S . equisimilis**. This is apparently a general feature of the **streptokinases** of this species. Sequencing of the skc gene from 3 of these strains indicated that the protein sequence of the protein is highly conserved within the species. (32 ref)

L6 ANSWER 6 OF 29 SCISEARCH COPYRIGHT (c) 2004 The Thomson Corporation. on STN
ACCESSION NUMBER: 97:555242 SCISEARCH
THE GENUINE ARTICLE: XL484
TITLE: The LppC gene of Streptococcus equisimilis encodes a lipoprotein that is homologous to the e(P4) outer membrane protein from Haemophilus influenzae
AUTHOR: Gase K; Liu G W; Bruckmann A; Steiner K; Ozegowski J; Malke H (Reprint)
CORPORATE SOURCE: UNIV JENA, INST MOL BIOL, WINZERLAER STR 10, D-07745 JENA, GERMANY (Reprint); UNIV JENA, INST MOL BIOL, D-07745 JENA, GERMANY
COUNTRY OF AUTHOR: GERMANY
SOURCE: MEDICAL MICROBIOLOGY AND IMMUNOLOGY, (JUN 1997) Vol. 186, No. 1, pp. 63-73.
Publisher: SPRINGER VERLAG, 175 FIFTH AVE, NEW YORK, NY 10010.
ISSN: 0300-8584.
DOCUMENT TYPE: Article; Journal
FILE SEGMENT: LIFE
LANGUAGE: English
REFERENCE COUNT: 39
ABSTRACT IS AVAILABLE IN THE ALL AND IALL FORMATS
AB We report the **cloning**, sequencing, and analysis of a novel chromosomal gene of Streptococcus equisimilis strain H46A that codes for a membrane lipoprotein, designated LppC. The lppC gene is located 3' adjacent to, and co-oriented with, the unrelated gapC gene that encodes

the previously characterized glyceraldehyde-3-phosphate dehydrogenase. Sequencing of lppC revealed an 855-bp open reading frame that predicted a 32.4-kDa polypeptide possessing a potential lipoprotein signal sequence and modification site (VTGC). Signal sequence processing of LppC synthesized in the homologous host or **expressed** from plasmid pLPP2 in Escherichia coli was sensitive to globomycin, a selective inhibitor of lipoprotein-specific signal peptidase II. Subcellular localization of LppC using polyclonal antibodies raised to the hexahistidyl-tagged protein proved LppC to be tightly associated with the cytoplasmic membrane of **S. equisimilis** and with the outer membrane of E. coli JM109 (pLPP2). Southern, Northern and Western analyses indicated that Ipl, was conserved in S. pyogenes, and transcribed independently of gap as monocistronic 0.9-kb mRNA from a sigma(70)-like consensus promoter. Database searches found homology of LppC to the hel gene-encoded outer membrane protein e (P4) from Haemophilus influenzae to which it exhibits 58% sequence similarity. However, unlike the hel gene, lppC was unable to complement hemA mutants of E. coli for growth on hemin as sole porphyrin source in aerobic conditions. Furthermore, neither the wild type nor an lppC insertion mutant of **S. equisimilis** could grow on hemin in iron-limited medium. These results, together with findings indicating that **S. equisimilis** H46A had no absolute requirement for iron, led us to conclude that lppC, in contrast to hel, is not involved in hemin utilization and has yet to be assigned a function.

L6 ANSWER 7 OF 29 LIFESCI COPYRIGHT 2004 CSA on STN
ACCESSION NUMBER: 96:44932 LIFESCI
TITLE: Functional analysis of a relA/spoT gene homolog from Streptococcus equisimilis
AUTHOR: Mechold, U.; Cashel, M.; Steiner, K.; Gentry, D.; Malke, H.
CORPORATE SOURCE: Inst. Molecular Biol., Jena Univ., Winzerlaer Str. 10, D-07745 Jena, Germany
SOURCE: J. BACTERIOL., (1996) vol. 178, no. 5, pp. 1404-1411. ISSN: 0021-9193.
DOCUMENT TYPE: Journal
FILE SEGMENT: J; G
LANGUAGE: English
SUMMARY LANGUAGE: English

AB We examined the functional attributes of a gene encountered by sequencing the **streptokinase** gene region of Streptococcus equisimilis H46A. This gene, originally called rel, here termed rel sub( )**S. equisimilis**, is homologous to two related Escherichia coli genes, spoT and relA, that function in the metabolism of guanosine 5',3'-polyphosphates [(p)ppGpp]. Studies with a variety of E. coli mutants led us to deduce that the highly **expressed** rel sub( )**S. equisimilis** gene encodes a strong (p)ppGppase and a weaker (p)ppGpp synthetic activity, much like the spoT gene, with a net effect favoring degradation and no complementation of the absence of the relA gene. We verified that the Rel sub( )**S. equisimilis** protein, purified from an E. coli relA spoT double mutant, catalyzed a manganese-activated (p)ppGpp 3'-pyrophosphohydrolase reaction similar to that of the SpoT enzyme. This Rel sub( )**S. equisimilis** protein preparation also weakly catalyzed a ribosome-independent synthesis of (p)ppGpp by an ATP to GTP 3'-pyrophosphoryltransferase reaction when degradation was restricted by the absence of manganese ions. An analogous activity has been deduced for the SpoT protein from genetic evidence. In addition, the Rel sub( )**S. equisimilis** protein displays immunological cross-reactivity with polyclonal antibodies specific for SpoT but not for RelA. Despite assignment of rel sub( ) **S. equisimilis** gene function in E. coli as being similar to that of the native spoT gene, disruptions of rel sub( )**S. equisimilis** in **S. equisimilis** abolish the parental (p)ppGpp accumulation response to amino acid starvation in a manner expected for relA mutants rather than spoT mutants.

L6 ANSWER 8 OF 29 MEDLINE on STN DUPLICATE 5
ACCESSION NUMBER: 96200111 MEDLINE
DOCUMENT NUMBER: PubMed ID: 8631718
TITLE: Functional analysis of a relA/spoT gene homolog from Streptococcus equisimilis.
AUTHOR: Mechold U; Cashel M; Steiner K; Gentry D; Malke H
CORPORATE SOURCE: Institute for Molecular Biology, Jena University, Germany.
SOURCE: Journal of bacteriology, (1996 Mar) 178 (5) 1401-11.
Journal code: 2985120R. ISSN: 0021-9193.
PUB. COUNTRY: United States
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 199607
ENTRY DATE: Entered STN: 19960715
Last Updated on STN: 19970203
Entered Medline: 19960703

AB We examined the functional attributes of a gene encountered by sequencing the **streptokinase** gene region of Streptococcus equisimilis H46A. This gene, originally called rel, here termed relS. equisimilis, is homologous to two related Escherichia coli genes, spoT and relA, that function in the metabolism of guanosine 5',3'-polyphosphates [(p)ppGpp]. Studies with a variety of E. coli mutants led us to deduce that the highly **expressed** rel **S. equisimilis** gene encodes a strong (p)ppGppase and a weaker (p)ppGpp synthetic activity, much like the spoT gene, with a net effect favoring degradation and no complementation of the absence of the relA gene. We verified that the Rel **S. equisimilis** protein, purified from an E. coli relA spoT double mutant, catalyzed a manganese-activated (p)ppGpp 3'-pyrophosphohydrolase reaction similar to that of the SpoT enzyme. This Rel **S. equisimilis** protein preparation also weakly catalyzed a ribosome-independent synthesis of (p)ppGpp by an ATP to GTP 3'-pyrophosphoryltransferase reaction when degradation was restricted by the absence of manganese ions. An analogous activity has been deduced for the SpoT protein from genetic evidence. In addition, the Rel **S. equisimilis** protein displays immunological cross-reactivity with polyclonal antibodies specific for SpoT but not for RelA. Despite assignment of rel **S. equisimilis** gene function in E. coli as being similar to that of the native spoT gene, disruptions of rel **S. equisimilis** in **S. equisimilis** abolish the parental (p)ppGpp accumulation response to amino acid starvation in a manner expected for relA mutants rather than spoT mutants.

L6 ANSWER 9 OF 29 MEDLINE on STN DUPLICATE 6
ACCESSION NUMBER: 96396845 MEDLINE
DOCUMENT NUMBER: PubMed ID: 8803948
TITLE: Structural dissection and functional analysis of the complex promoter of the **streptokinase** gene from Streptococcus equisimilis H46A.
AUTHOR: Grafe S; Ellinger T; Malke H
CORPORATE SOURCE: Institute for Molecular Biology, Jena University, Germany.
SOURCE: Medical microbiology and immunology, (1996 May) 185 (1) 11-7.
Journal code: 0314524. ISSN: 0300-8584.
PUB. COUNTRY: GERMANY: Germany, Federal Republic of
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 199701
ENTRY DATE: Entered STN: 19970219
Last Updated on STN: 19970219
Entered Medline: 19970131

AB The overlapping tandem promoters of the **streptokinase** gene, P1

and P2, identified previously by S1 nuclease transcript mapping were functionally dissected by mutagenesis of their -10 regions and fused transcriptionally with or without the 202-bp upstream region (USR) to the luciferase reporter gene (luc) from Photinus pyralis to analyze the contribution of the different sequence elements to promoter activity in Escherichia coli and the homologous Streptococcus equisimilis strain H46A. In E. coli, virtually the entire promoter activity derived from the upstream promoter P1. In **S. equisimilis**, luc **expression** increased in the following order of the involved sequence elements: P2 approximately equal to P2 + USR < P1 < P1 + P2 < P1 + USR < P1 + P2 + USR. This shows that (1) in the homologous system, P1 and P2 alone are extremely weak, (2) in the USR-less arrangement, only the combined core promoters have substantial activity, and (3) the USR stimulates only P1 and the combination of P1 + P2. Thus, the tandem promoters presumably function by mutual contributary action and their full activity strongly depends on the AT-rich and statically bent upstream region. The distinctive feature determining the strength of P1 in both hosts appears to be its extended -10 region which matches the consensus TRTGN established for strong S. pneumoniae and Bacillus subtilis promoters.

L6 ANSWER 10 OF 29 HCAPLUS COPYRIGHT 2004 ACS on STN
ACCESSION NUMBER: 2000:433656 HCAPLUS
DOCUMENT NUMBER: 133:27355
TITLE: **Cloning** and **expression** of Streptococococcus H46 **streptokinase** gene
INVENTOR(S): Cho, Jung-Myong; Park, Yong-U.
PATENT ASSIGNEE(S): LG Chemical Co., Ltd., S. Korea
SOURCE: Repub. Korea, No pp. given
CODEN: KRXXFC
DOCUMENT TYPE: Patent
LANGUAGE: Korean
FAMILY ACC. NUM. COUNT: 1
PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
KR 9512901	B1	19951023	KR 1992-17406	19920924

PRIORITY APPLN. INFO.: KR 1992-17406 19920924

AB The **cloning** of **streptokinase** gene of Streptococcus H46 consists of PCR with primers and **cloning** the gene into the PstI-NdeI site of plasmid ptrp322H-HGH (KFCC 10067) to get ptrpH-SK (ATCC 68884). The DNA sequence of Streptococcus H46 **streptokinase** has 92.2-98.8% homol. to SKC, SKG, and SKA. Streptococcus H46 is also designated **S. equisimilis** ATCC 35556.

L6 ANSWER 11 OF 29 MEDLINE on STN DUPLICATE 7
ACCESSION NUMBER: 95342169 MEDLINE
DOCUMENT NUMBER: PubMed ID: 7616967
TITLE: Complex transcriptional control of the **streptokinase** gene of Streptococcus equisimilis H46A.
AUTHOR: Gase K; Ellinger T; Malke H
CORPORATE SOURCE: Institute for Molecular Biology, Jena University, Germany.
SOURCE: Molecular & general genetics : MGG, (1995 Jun 25) 247 (6) 749-58.
Journal code: 0125036. ISSN: 0026-8925.
PUB. COUNTRY: GERMANY: Germany, Federal Republic of
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 199508
ENTRY DATE: Entered STN: 19950905
Last Updated on STN: 19950905

Entered Medline: 19950822

AB On the Streptococcus equisimilis H46A chromosome, the divergent coding sequences of the genes for the plasminogen activator **streptokinase** (skc) and a leucine-rich protein (lrp), the function of which is unknown, are separated by a 328 bp intrinsically bent DNA region rich in AT tracts. To begin to understand the **expression** control of these two genes, we mapped their transcriptional initiation sites by S1 nuclease analysis and studied the influence of the bent intergenic region on promoter strength, using promoter-reporter gene fusions of skc' and lrp' to 'lacZ from Escherichia coli. The major transcriptional start sites, in both **S. equisimilis** and E. coli, mapped 22 bases upstream of the ATG start site of lrp (G), and 24 and 32 bases upstream of the translational initiation codon of skc (A and G, respectively), indicating the existence of two overlapping canonical skc promoters arranged in tandem on opposite faces of the helix. The reporter gene fusions were **cloned** in E. coli on a vector containing a 1.1 kb fragment of the **S. equisimilis** dexB gene, thus allowing promoter strength to be measured in multiple plasmid-form copies in the heterologous host and in single-copy genomic form following integration into the skc region of the homologous host. In **S. equisimilis**, skc'-'lacZ was **expressed** about 200-fold more strongly than the corresponding lrp'-'lacZ fusion. In contrast, in E. coli, the corresponding levels of **expression** differed by only about 11-fold. Deletion of the 202 bp bent region upstream of the skc and lrp core promoters caused a 13-fold decrease in skc promoter activity in **S. equisimilis** but did not alter lrp promoter strength in this host. In contrast, when studied in E. coli, this deletion did not alter the strength of the skc-double promoter and even increased by 2.4- to 3-fold the activity of the lrp promoter. This comparative promoter analysis shows that skc has a complex promoter structure, the activity of which in the homologous genomic environment specifically depends on sequences upstream of the two core promoters. Thus, the skc promoter structure resembles that of an array of promoters involved in a transcriptional switch; however, the nature of the potential switch factor(s) remains unknown.

L6 ANSWER 12 OF 29 MEDLINE on STN DUPLICATE 8

ACCESSION NUMBER: 95157528 MEDLINE
DOCUMENT NUMBER: PubMed ID: 7531815
TITLE: Transcription termination of the **streptokinase** gene of Streptococcus equisimilis H46A: bidirectionality and efficiency in homologous and heterologous hosts.
AUTHOR: Steiner K; Malke H
CORPORATE SOURCE: Institute for Molecular Biology, Jena University, Germany.
SOURCE: Molecular & general genetics : MGG, (1995 Feb 6) 246 (3) 374-80.
Journal code: 0125036. ISSN: 0026-8925.
PUB. COUNTRY: GERMANY: Germany, Federal Republic of
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 199503
ENTRY DATE: Entered STN: 19950322
Last Updated on STN: 19960129
Entered Medline: 19950316

AB In Streptococcus equisimilis H46A, a hypersymmetrical transcription terminator with bidirectional activity was localized between the translational termination codons of the **streptokinase** gene, skc, and the rel-orf1 genes. These two transcription units are oriented towards each other, and under normal conditions the skc mRNA level exceeds that of the rel-orf1 genes by a factor of at least 1000. Reporter vectors based on the promoterless cat gene were constructed by transcriptional fusion of skc to cat, such that the region between the two genes contained the terminator in skc orientation or in rel-orf1 orientation.

Additionally, skc and cat were fused directly, with deletion of the terminator. The reporter vectors were designed to be capable of being studied either as multicopy plasmids in Escherichia coli or in single copy following integration, via skc, into the **S. equisimilis** chromosome. Chloramphenicol acetyl transferase (CAT) activity assays in conjunction with determination of chloramphenicol resistance levels and Northern hybridization analysis showed that the terminator is active in either host and orientation. However, termination efficiency was host dependent, with high terminator strength being observed in the homologous streptococcal background and appreciable readthrough occurring in E. coli. The extent of transcriptional readthrough was dependent upon terminator orientation, with termination being more efficient in rel-orf1 polarity. The results suggest that, in **S. equisimilis**, transcription of both skc and rel-orf1 is efficiently terminated by a common signal, and that these genes are largely protected from convergent transcription, which otherwise would seem to be particularly detrimental to the weakly **expressed** rel-orf1 genes.

L6 ANSWER 13 OF 29 MEDLINE on STN DUPLICATE 9
ACCESSION NUMBER: 96154934 MEDLINE
DOCUMENT NUMBER: PubMed ID: 8577315
TITLE: Conservation of the organization of the **streptokinase** gene region among pathogenic streptococci.
AUTHOR: Frank C; Steiner K; Malke H
CORPORATE SOURCE: Institute for Molecular Biology, Jena University, Germany.
SOURCE: Medical microbiology and immunology, (1995 Oct) 184 (3) 139-46.
Journal code: 0314524. ISSN: 0300-8584.
PUB. COUNTRY: GERMANY: Germany, Federal Republic of
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
OTHER SOURCE: GENBANK-X72832
ENTRY MONTH: 199603
ENTRY DATE: Entered STN: 19960321
Last Updated on STN: 19960321
Entered Medline: 19960313

AB Using ten gene-specific probes from the **cloned** and sequenced **streptokinase** gene (skc) region (8,931 bp) of Streptococcus equisimilis H46A, a human serogroup C strain, the conservation of these genes and their linkage relationships were studied by Southern hybridization in pathogenic streptococci differing taxonomically, serologically, in regard to their host range, and in the class of plasminogen activator produced. The results indicate that in S. pyogenes (strains A374, NZ131 and SF130/13) and a human group G strain (G19,908) both gene content and gene order as determined for H46A (dexB-abc-lrp-skc-orf1-rel) are preserved. The same is true of an equine **S. equisimilis** isolate (87-542-W), the **streptokinase** gene of which has been shown to hybridize detectably with skc, a result at variance with that obtained previously by others. In contrast, the chromosomal DNA of three S. uberis strains (0140J, C198, C216) of bovine origin, two of which produced a plasminogen activator different from **streptokinase**, hybridized only with dexB-, abc- and rel-specific probes, and the homologues of these genes appeared to lie close to each other. The maintenance of the organization of the **streptokinase** gene region in strains differing in overall chromosomal character suggests that this gene arrangement is of selective advantage.

L6 ANSWER 14 OF 29 HCAPLUS COPYRIGHT 2004 ACS on STN
ACCESSION NUMBER: 1994:186995 HCAPLUS
DOCUMENT NUMBER: 120:186995
TITLE: Inactivation of the **streptokinase** gene

prevents Streptococcus equisimilis H46A from acquiring cell-associated plasmin activity in the presence of plasminogen

AUTHOR(S): Malke, Horst; Mechold, Undine; Gase, Klaus; Gerlach, Dieter

CORPORATE SOURCE: Inst. Mol. Biol., Jena Univ., Jena, D-07745, Germany

SOURCE: FEMS Microbiology Letters (1994), 116(1), 107-12
CODEN: FMLED7; ISSN: 0378-1097

DOCUMENT TYPE: Journal

LANGUAGE: English

AB The **streptokinase** gene of **S. equisimilis** H46 was inactivated by plasmid insertion mutagenesis to study the relation between elaboration of **streptokinase** and acquisition of cell-associated plasmin activity after incubation of wild-type and mutant cells in media containing plasminogen or plasmin. H46A binds both the zymogen and active enzyme, generates surface-associated plasmin activity in the presence of plasminogen when producing **streptokinase**, and **expresses** its plasmin(ogen) receptor(s) independently of a functional **streptokinase** gene. At least part of the plasmin(ogen) binding capacity may be due to the glyceraldehyde-3-phosphate dehydrogenase type of receptor mol., as judged by the detection of the corresponding gene.

L6 ANSWER 15 OF 29 HCAPLUS COPYRIGHT 2004 ACS on STN

ACCESSION NUMBER: 1992:441935 HCAPLUS

DOCUMENT NUMBER: 117:41935

TITLE: **Cloning** and **expression** of **streptokinase** gene of C-type Streptococcus equisimilis

PATENT ASSIGNEE(S): Centro de Ingenieria Genetica y Biotecnologia (CIGB), Cuba

SOURCE: Jpn. Kokai Tokkyo Koho, 12 pp.
CODEN: JKXXAF

DOCUMENT TYPE: Patent

LANGUAGE: Japanese

FAMILY ACC. NUM. COUNT: 2

PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
JP 04030794	A2	19920203	JP 1990-201600	19900731
JP 3127298	B2	20010122		
EP 489201	A1	19920610	EP 1990-201930	19900717
EP 489201	B1	19951115		
R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE				
AT 130369	E	19951215	AT 1990-201930	19900717
ES 2081909	T3	19960316	ES 1990-201930	19900717
US 5296366	A	19940322	US 1991-703778	19910522
AU 644657	B2	19931216	AU 1991-78101	19910531
RU 2107726	C1	19980327	RU 1991-5001280	19910717
PRIORITY APPLN. INFO.:			CU 1990-90 A	19900523
			SU 1991-5001280 A	19910717

AB The **streptokinase** (I) gene SKC-2 ,with/without signal sequence, is **cloned** from C-type **S. equisimilis** ATCC-9542 by the polymerase chain reaction method and **expressed** in Escherichia coli and yeast for com. manufacture of I. Genomic DNA of the C-type **S. equisimilis** was isolated by the standard method and amplified with primers derived from the nucleotide sequence of SKC to get I gene with/without signal sequence. **Expression** of the I gene in E. coli and Pichia pastoris MP-36 mutant were shown. The production of I with these microorganisms were ≥350 mg/L and ≥1.2 g/L, resp.

L6 ANSWER 16 OF 29 HCAPLUS COPYRIGHT 2004 ACS on STN

ACCESSION NUMBER: 1992:646505 HCAPLUS
DOCUMENT NUMBER: 117:246505
TITLE: **Streptokinase** mutation affecting skc **expression** in homologous and heterologous hosts
AUTHOR(S): Mechold, U.; Muller, J.; Malke, H.
CORPORATE SOURCE: Cent. Inst. Microbiol. Exp. Ther., Jena, D-6900, Germany
SOURCE: Zentralblatt fuer Bakteriologie, Supplement (1992), 22(New Perspect. Streptococci Streptococcal Infect.), 336-8
CODEN: ZBASE2; ISSN: 0941-018X
DOCUMENT TYPE: Journal
LANGUAGE: English

AB Mutations affecting the level of **streptokinase** gene skc **expression** and/or secretion in homologous and heterologous hosts are phys. characterized. The principal classes of mutations produced included skc deletions, IS element insertions, and skc duplications. The deletion events, represented by mutations Δ(skc)-247 and Δ(skc)-305 present in plasmids pMM247 and pMM305, resp., removed a tetrapeptide (F10-L13 or L12-A15) from the hydrophobic core of the Skc signal sequence. These mutations, reduced the size, hydrophobicity and predicted alpha-helicity of the central region of the signal sequence. The corresponding plasmids, upon transformation into E. coli and P. mirabilis L-forms, substantially increased the level of Skc **expression** in either host. In E. coli, they also facilitated the export of mature Skc into the culture medium. In the gram-pos. hosts, skc **expression** was less dramatically affected; however, the proportion of Skc activity found in the culture medium was significantly decreased when compared to the extracellular activity resulting from wild type skc. IS1 insertion did not alter the primary structure of the promoter but displaced in upward direction, by 768 bp, a static DNA bending locus having its center some 140 bp upstream of the -35 region in wild type DNA. When studied with plasmid pMM697, this insertion event resulted in severely decreased Skc **expression** in all hosts but, expectedly, did not affect Skc secretability. Gene skc duplication in the chromosome of the homologous producer strain, **S. equisimilis** H46A, was achieved by a single crossover event between the chromosomes and an integrateable Skc plasmid, pSM752, in the region of shared homol. As judged by Southern hybridization, cells transiently supporting the replication of pSM752 gave rise to a stable erythromycin-resistant **clone** designated H46SM which was plasmid-free and produced Skc at levels approx. twice as high as the wild type.

L6 ANSWER 17 OF 29 MEDLINE on STN DUPLICATE 10
ACCESSION NUMBER: 92039051 MEDLINE
DOCUMENT NUMBER: PubMed ID: 1937032
TITLE: Isolation, sequence and **expression** in Escherichia coli, Bacillus subtilis and Lactococcus lactis of the DNase (streptodornase)-encoding gene from Streptococcus equisimilis H46A.
AUTHOR: Wolinowska R; Ceglowski P; Kok J; Venema G
CORPORATE SOURCE: Department of Pharmaceutical Microbiology, Medical Academy, Warsaw, Poland.
SOURCE: Gene, (1991 Sep 30) 106 (1) 115-9.
Journal code: 7706761. ISSN: 0378-1119.
PUB. COUNTRY: Netherlands
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
OTHER SOURCE: GENBANK-M59725; GENBANK-M59726; GENBANK-M59727; GENBANK-M59728; GENBANK-M63990; GENBANK-S61507; GENBANK-S63856; GENBANK-S63863; GENBANK-S65020; GENBANK-S65060; GENBANK-X17241

ENTRY MONTH: 199112
ENTRY DATE: Entered STN: 19920124
Last Updated on STN: 19920124
Entered Medline: 19911223

AB A partial library of BclI-generated chromosomal DNA fragments from Streptococcus equisimilis H64A (Lancefield Group C) was constructed in Escherichia coli. **Clones** displaying either **streptokinase** or deoxyribonuclease (streptodornase; SDC) activities were isolated. The gene (sdc) **expressing** the SDC activity was allocated on the 1.1-kb AccI DNA subfragment. Sequence analysis of this DNA fragment revealed the presence of one open reading frame, which could encode a protein of 36.8 kDa. The N-terminal portion of the deduced protein exhibited features characteristic of prokaryotic signal peptides. The sdc gene was **expressed** in E. coli, Bacillus subtilis and Lactococcus lactis. As observed for **S. equisimilis**, in the heterologous Gram + hosts, at least part of the SDC protein was secreted into the medium.

L6 ANSWER 18 OF 29 SCISEARCH COPYRIGHT (c) 2004 The Thomson Corporation. on STN
ACCESSION NUMBER: 91:636602 SCISEARCH
THE GENUINE ARTICLE: GQ068
TITLE: ISOLATION, SEQUENCE AND **EXPRESSION** IN ESCHERICHIA-COLI, BACILLUS-SUBTILIS AND LACTOCOCCUS-LACTIS OF THE DNASE (STREPTODORNASE)-ENCODING GENE FROM STREPTOCOCCUS-EQUISIMILIS H46A
AUTHOR: WOLINOWSKA R; CEGLOWSKI P (Reprint); KOK J; VENEMA G
CORPORATE SOURCE: MED ACAD WARSAW, DEPT PHARMACEUT MICROBIOL, OCZKI 3, PL-02007 WARSAW, POLAND; UNIV GRONINGEN, INST GENET, 9700 AB GRONINGEN, NETHERLANDS
COUNTRY OF AUTHOR: POLAND; NETHERLANDS
SOURCE: GENE, (1991) Vol. 106, No. 1, pp. 115-119.
DOCUMENT TYPE: Note; Journal
FILE SEGMENT: LIFE
LANGUAGE: ENGLISH
REFERENCE COUNT: 32

ABSTRACT IS AVAILABLE IN THE ALL AND IALL FORMATS

AB A partial library of BclI-generated chromosomal DNA fragments from Streptococcus equisimilis H64A (Lancefield Group C) was constructed in Escherichia coli. **Clones** displaying either **streptokinase** or deoxyribonuclease (streptodornase; SDC) activities were isolated. The gene (sdc) **expressing** the SDC activity was allocated on the 1.1-kb AccI DNA subfragment. Sequence analysis of this DNA fragment revealed the presence of one open reading frame, which could encode a protein of 36.8 kDa. The N-terminal portion of the deduced protein exhibited features characteristic of prokaryotic signal peptides. The sdc gene was **expressed** in E. coli, Bacillus subtilis and Lactococcus lactis. As observed for **S. equisimilis**, in the heterologous Gram+ hosts, at least part of the SDC protein was secreted into the medium.

L6 ANSWER 19 OF 29 HCAPLUS COPYRIGHT 2004 ACS on STN
ACCESSION NUMBER: 1991:179756 HCAPLUS
DOCUMENT NUMBER: 114:179756
TITLE: Manufacture of serotype c **streptokinase** with **recombinant** Streptococcus equisimilis
INVENTOR(S): Mueller, Joerg; Malke, Horst
PATENT ASSIGNEE(S): Akademie der Wissenschaften der DDR, Ger. Dem. Rep.
SOURCE: Ger. (East), 12 pp.
CODEN: GEXXA8
DOCUMENT TYPE: Patent
LANGUAGE: German
FAMILY ACC. NUM. COUNT: 1
PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
DD 284898	A5	19901128	DD 1989-332866	19890921
PRIORITY APPLN. INFO.:			DD 1989-332866	19890921

AB Serotype c **streptokinase** is manufactured by **S. equisimilis** transformed with a plasmid containing the **S. equisimilis** skc gene and a selectable marker, preferably the erythromycin resistance gene. The plasmid becomes incorporated into the microbial genome by recombination to double the skc gene copy number to two. Submerged cultivation of the transformant results in the enzyme being secreted into the medium in quantities .apprx.2-fold greater than those secreted by the wild-type strain.

L6 ANSWER 20 OF 29 HCAPLUS COPYRIGHT 2004 ACS on STN
ACCESSION NUMBER: 1990:625400 HCAPLUS
DOCUMENT NUMBER: 113:225400
TITLE: Duplication of the **streptokinase** gene in the chromosome of Streptococcus equisimilis H46A
AUTHOR(S): Mueller, Joerg; Malke, Horst
CORPORATE SOURCE: Acad. Sci. GDR, Cent. Inst. Microbiol. Exp. Ther., Jena, DDR-6900, Ger. Dem. Rep.
SOURCE: FEMS Microbiology Letters (1990), 72(1-2), 75-8
CODEN: FMLED7; ISSN: 0378-1097
DOCUMENT TYPE: Journal
LANGUAGE: English

AB The erythromycin resistance plasmid pSM752 carrying the **cloned streptokinase** gene, skc, was introduced by protoplast transformation into **S. equisimilis** H46A from which skc was originally **cloned**. Cells transiently supporting the replication of pSM752 gave rise to an erythromycin-resistant **clone** designated H46SM which was plasmid free and produced **streptokinase** at levels approx. twice as high as the wild type. Southern hybridization of total cell DNA with an skc-containing probe provided evidence for the duplication of the skc gene in the H46SM chromosome. The results, which have some bearing on industrial **streptokinase** production, can be best explained by a single cross-over event between the chromosome and the plasmid in the region of shared homol. leading to the integration of pSM752 in a Campbell-like manner.

L6 ANSWER 21 OF 29 HCAPLUS COPYRIGHT 2004 ACS on STN
ACCESSION NUMBER: 1988:88986 HCAPLUS
DOCUMENT NUMBER: 108:88986
TITLE: **Expression** of **streptokinase** gene of Streptococcus in Pichia pastoris
INVENTOR(S): Hagenson, Mary Jane; Stroman, David Womack
PATENT ASSIGNEE(S): Phillips Petroleum Co. , USA
SOURCE: Eur. Pat. Appl., 27 pp.
CODEN: EPXXDW
DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 1
PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
EP 248227	A1	19871209	EP 1987-106614	19870507
R: AT, BE, CH, DE, ES, FR, GB, GR, IT, LI, LU, NL, SE				
ZA 8702534	A	19871125	ZA 1987-2534	19870408
AU 8771390	A1	19871112	AU 1987-71390	19870410
AU 592862	B2	19900125		
JP 62296881	A2	19871224	JP 1987-109620	19870502
NO 8701886	A	19871109	NO 1987-1886	19870506
DK 8702335	A	19871109	DK 1987-2335	19870507

FI 8702031	A	19871109	FI 1987-2031	19870507
BR 8702337	A	19880217	BR 1987-2337	19870507
DD 257646	A5	19880622	DD 1987-302541	19870507

PRIORITY APPLN. INFO.: US 1986-860960 A 19860508

AB The gene for **streptokinase** of Streptococcus equisimilis is **cloned** and **expressed** in Pichia pastoris. Plasmid pHTskc25 was constructed containing the coding sequence (minus the signal sequence) for **S. equisimilis streptokinase** under the control of the alc. oxidase gene promoter of P. pastoris. P. pastoris Transformed with the plasmid and grown in MeOH-containing medium produced 16 units **streptokinase**/O.D. cells.

L6 ANSWER 22 OF 29 HCAPLUS COPYRIGHT 2004 ACS on STN

ACCESSION NUMBER: 1986:455636 HCAPLUS
DOCUMENT NUMBER: 105:55636
TITLE: The **streptokinase** gene: **cloning**, sequencing and **expression** in new hosts
AUTHOR(S): Malke, Horst
CORPORATE SOURCE: Zentralinst. Mikrobiol., Dtsch. Akad. Wiss., Jena, Ger. Dem. Rep.
SOURCE: Zeitschrift fuer Klinische Medizin (1985) (1986), 41(7), 502-4
CODEN: ZKMEEF; ISSN: 0233-1608
DOCUMENT TYPE: Journal
LANGUAGE: German

AB The **streptokinase** (I) [9002-01-1] gene (skc) of Streptococcus equisimilis H46A was **cloned** in Escherichia coli using vector λL47. One of the **recombinant clones** was used to subclone skc in E. coli plasmid vectors. Plasmids pMF2 (10.4 kilobases, composed of pACYC184 plus a 6.4-kilobase EcoRI fragment) and pMF5 (6.9 kilobases, with a 2.5-kilobase fragment in the PstI site of pBR322) determined I formation in E. coli; **expression** of skc was independent of its orientation, indicating that the complete gene, together with its control elements, was present. The 2.5-kilobase PstI fragment of pMF5 was isolated and sequenced in the M13 system. Of 2568 base pairs, the largest open reading frame consisted of 1320 base pairs coding for prestreptokinase, corresponding to I plus its 26-amino acid leader sequence. **Expression** of skc was attained in S. sanguis after transformation with the shuttle vector pSM752. In fermentation expts., I production rates of 1500 U/mL were attained, which was below the levels obtained with **S. equisimilis**. Use of pSM752 for similar transformation of Bacillus subtilis is briefly discussed.

L6 ANSWER 23 OF 29 BIOSIS COPYRIGHT (c) 2004 The Thomson Corporation. on STN DUPLICATE 11

ACCESSION NUMBER: 1987:277792 BIOSIS
DOCUMENT NUMBER: PREV198784018831; BA84:18831
TITLE: MOLECULAR **CLONING** OF **STREPTOKINASE** GENE FROM STREPTOCOCCUS-EQUISIMILIS AND ITS **EXPRESSION** IN ESCHERICHIA-COLI.
AUTHOR(S): ROH D C [Reprint author]; KIM J H; PARK S K; LEE J W; BYRUN S M
CORPORATE SOURCE: DEP BIOLOGICAL SCIENCE AND ENGINEERING, KOREA ADVANCED INST SCIENCE AND TECHNOLOGY KAIST , PO BOX 150 CHONGRYANG, SEOUL 131, KOREA
SOURCE: Korean Biochemical Journal, (1986) Vol. 19, No. 4, pp. 391-398.
CODEN: KBCJAK. ISSN: 0368-4881.
DOCUMENT TYPE: Article
FILE SEGMENT: BA
LANGUAGE: ENGLISH
ENTRY DATE: Entered STN: 19 Jun 1987
Last Updated on STN: 19 Jun 1987

AB The streptococcal genomic DNA digested with Pst I was **cloned** in

E. coli HB101. The overlay technique of casein/plasminogen was used to screen the **clones** for **recombinants** carrying the **streptokinase** gene. The insert size of the plasmid carrying the **streptokinase** gene was a 2.5, 4.3, and 5.8 Kb, respectively. The restriction maps of all three hybrid plasmids were constructed by digestion with Pst I, Pvu II, Sal I, Hind III, Ava I, BamH I, and Cla I. For the identification of **cloned** gene, **streptokinase** was highly purified from **S. equisimilis** by the methods of gel chromatography and isoelectric focusing and rabbits were immunized with this purified **streptokinase**. Several lines of evidence, including proof obtained by the immunodiffusion technique, established that the enzyme from E. coli was identical to that from **S. equisimilis**. In the E. coli cell culture, we found the activity of **streptokinase** in all three principal locations of the cell. More than 50% were existed in the intracellular space.

L6 ANSWER 24 OF 29 HCAPLUS COPYRIGHT 2004 ACS on STN
ACCESSION NUMBER: 1986:220084 HCAPLUS
DOCUMENT NUMBER: 104:220084
TITLE: **Expression** of the **streptokinase** gene from Streptococcus equisimilis in Bacillus subtilis
AUTHOR(S): Klessen, Christian; Malke, Horst
CORPORATE SOURCE: Cent. Inst. Microbiol. Exp. Ther., Acad. Sci. GDR, Jena, 6900, Ger. Dem. Rep.
SOURCE: Journal of Basic Microbiology (1986), 26, 75-81
CODEN: JBMIEQ; ISSN: 0233-111X
DOCUMENT TYPE: Journal
LANGUAGE: English
AB The previously **cloned** and sequenced **streptokinase** [9002-01-1] gene (skc) from **S. equisimilis** H46A was inserted into plasmid vectors capable of replication in B. subtilis. The skc gene was **expressed** by use of its own transcription and translation signals which appeared to meet the stringent requirements of B. subtilis for efficient foreign gene **expression**. The secreted **streptokinase** activity began to decline toward the end of the exponential growth phase suggesting that B. subtilis exoproteases hydrolyzed and inactivated the foreign protein.

L6 ANSWER 25 OF 29 HCAPLUS COPYRIGHT 2004 ACS on STN
ACCESSION NUMBER: 1985:482518 HCAPLUS
DOCUMENT NUMBER: 103:82518
TITLE: Nucleotide sequence of the **streptokinase** gene from Streptococcus aquisimilis H46A
AUTHOR(S): Malke, Horst; Roe, Bruce; Ferretti, Joseph J.
CORPORATE SOURCE: Health Sci. Cent., Univ. Oklahoma, Oklahoma City, OK, 73190, USA
SOURCE: Gene (1985), 34(2-3), 357-62
CODEN: GENED6; ISSN: 0378-1119
DOCUMENT TYPE: Journal
LANGUAGE: English
AB The entire nucleotide sequence of a **cloned** 2568-base-pair (bp) PstI fragment from the genome of **S. equisimilis** H46A encoding the **streptokinase** [9002-01-1] gene (skc) was determined The longest open reading frame comprises 1320 bp which code for **streptokinase**. The protein is synthesized with a 26-amino acid residue N-terminal extension having properties characteristic of a signal peptide. Comparison of the deduced amino acid sequence with the available amino acid sequence of a com. **streptokinase** reveals minor structure differences. The nucleotide sequencing of skc does not support the hypothesis that the gene has evolved by duplication and fusion, as suggested by internal 2-fold amino acid homologies of its product. Furthermore, the skc gene sequence shows no extended regions homologous to the staphylokinase gene. Upstream from the skc gene, the putative skc

promoter and the ribosome-binding site sequence were identified; downstream from the coding region, inverted repeat sequences thought to function as transcription terminators were detected.

L6 ANSWER 26 OF 29 HCAPLUS COPYRIGHT 2004 ACS on STN
ACCESSION NUMBER: 1986:473464 HCAPLUS
DOCUMENT NUMBER: 105:73464
TITLE: Hybridization of a **cloned** group C streptococcal **streptokinase** gene with DNA from other streptococcal species
AUTHOR(S): Huang, T. T.; Malke, H.; Ferretti, J. J.
CORPORATE SOURCE: Health Sci. Cent., Univ. Oklahoma, Oklahoma City, OK, USA
SOURCE: Recent Adv. Streptococci Streptococcal Dis., Proc. Lancefield Int. Symp. Streptococci Streptococcal Dis., 9th (1985), Meeting Date 1984, 234-6. Editor(s): Kimura, Yoshitami; Kotani, Shozo; Shiokawa, Yuichi. Reedbooks: Bracknell, UK.
CODEN: 55BSAN
DOCUMENT TYPE: Conference
LANGUAGE: English
AB The previously **cloned streptokinase** [9002-01-1] gene (skc) of Streptococcus equisimilis and 2 subfragments were used as DNA hybridization probes to determine sequence homologies with other streptococcal species. The human pathogenic streptococci of strains A, C, and G were the only strains that had a pos. correlation between the ability to produce **streptokinase** and to hybridize with the gene skc DNA probe. In conjunction with other streptococcal DNA probes, such as streptolysin O, hyaluronidase, DNase, and erythrogenic toxins, the skc probe may be of diagnostic significance in the rapid identification of human pathogenic streptococci.

L6 ANSWER 27 OF 29 HCAPLUS COPYRIGHT 2004 ACS on STN
ACCESSION NUMBER: 1986:473709 HCAPLUS
DOCUMENT NUMBER: 105:73709
TITLE: **Cloning** of streptococcal genes with Streptococcus-Escherichia coli shuttle vector pSA3
AUTHOR(S): Dao, M. L.; Ferretti, J. J.
CORPORATE SOURCE: Health Sci. Cent., Univ. Oklahoma, Oklahoma City, OK, USA
SOURCE: Recent Adv. Streptococci Streptococcal Dis., Proc. Lancefield Int. Symp. Streptococci Streptococcal Dis., 9th (1985), Meeting Date 1984, 233-4. Editor(s): Kimura, Yoshitami; Kotani, Shozo; Shiokawa, Yuichi. Reedbooks: Bracknell, UK.
CODEN: 55BSAN
DOCUMENT TYPE: Conference
LANGUAGE: English
AB A shuttle vector, the chimeric plasmid pSA3, which can replicate in both E. coli and S. sanguis, was constructed. Chromosomal DNA from S. mutans was ligated into this plasmid and **cloned** in E. coli. Of 472 **clones** tested, 43 **clones expressed** S. mutans surface antigens. A **cloned S. equisimilis streptokinase** [9002-01-1] gene was inserted into plasmid pSA3 and then used to transform E. coli, S. sanguis, and S. mutans, all of which **expressed** the **cloned streptokinase** gene.

L6 ANSWER 28 OF 29 HCAPLUS COPYRIGHT 2004 ACS on STN
ACCESSION NUMBER: 1986:473707 HCAPLUS
DOCUMENT NUMBER: 105:73707
TITLE: **Cloned streptokinase** gene from Streptococcus equisimilis H46A
AUTHOR(S): Malke, H.; Ferretti, J. J.
CORPORATE SOURCE: Ger. Acad. Sci., Jena, Ger. Dem. Rep.

SOURCE: Recent Adv. Streptococci Streptococcal Dis., Proc. Lancefield Int. Symp. Streptococci Streptococcal Dis., 9th (1985), Meeting Date 1984, 221-2. Editor(s): Kimura, Yoshitami; Kotani, Shozo; Shiokawa, Yuichi. Reedbooks: Bracknell, UK.
CODEN: 55BSAN
DOCUMENT TYPE: Conference
LANGUAGE: English
AB The **streptokinase** [9002-01-1] gene skc of **S. equisimilis** was **cloned** in Escherichia coli with plasmid pBR322. **Expression** of gene skc was observed with both orientations of the gene, which indicated that its own promoter was present and was functional in E. coli. **Streptokinase** was excreted by the E. coli host. The gene contained a 1320-base-pair open reading frame which encodes 440 amino acids, including a signal peptide of 26 amino acids.

L6 ANSWER 29 OF 29 BIOSIS COPYRIGHT (c) 2004 The Thomson Corporation. on STN DUPLICATE 12
ACCESSION NUMBER: 1985:232658 BIOSIS
DOCUMENT NUMBER: PREV198579012654; BA79:12654
TITLE: **EXPRESSION** OF A **STREPTOKINASE** GENE FROM STREPTOCOCCUS-EQUISIMILIS IN STREPTOCOCCUS-SANGUIS.
AUTHOR(S): MALKE H [Reprint author]; GERLACH D; KOEHLER W; FERRETTI J J
CORPORATE SOURCE: ACAD SCI GDR, CENTRAL INST MICROBIOLOGY EXPERIMENTAL THERAPY, DDR-69 JENA, GDR
SOURCE: Molecular and General Genetics, (1984) Vol. 196, No. 2, pp. 360-363.
CODEN: MGGEAE. ISSN: 0026-8925.
DOCUMENT TYPE: Article
FILE SEGMENT: BA
LANGUAGE: ENGLISH
AB Using **recombinant** DNA techniques, one introduced a previously **cloned streptokinase** gene from **S. equisimilis** into the Challis strain of S. sanguis (group H). The gene was **expressed** in the new host under the control of its own promoter and the gene product had biological properties identical to authentic **streptokinase**. The MW of **cloned streptokinase** (42 K [kilodalton]) as **expressed** by S. sanguis was substantially lower than that of authentic **streptokinase** (47 K). Since the **cloned streptokinase** gene encoded a 47 K mature protein, the lowered MW of S. sanguis **streptokinase** may reflect posttranslational proteolytic cleavage, which leaves the biological activity of the gene product and its serological reactivity unimpaired.

=> d his

(FILE 'HOME' ENTERED AT 11:36:50 ON 10 DEC 2004)

FILE 'MEDLINE, EMBASE, BIOSIS, BIOTECHDS, SCISEARCH, HCAPLUS, NTIS, LIFESCI' ENTERED AT 11:37:16 ON 10 DEC 2004
L1 41164 S STREPTOKINASE?
L2 364 S "S. EQUISIMILIS"
L3 103 S L1 AND L2
L4 6828585 S CLON? OR EXPRESS? OR RECOMBINANT
L5 74 S L3 AND L4
L6 29 DUP REM L5 (45 DUPLICATES REMOVED)

=> e kuppusamy m/au
E1 1 KUPPUSAMY KAVITHA/AU
E2 2 KUPPUSAMY KAVITHA T/AU
E3 40 --> KUPPUSAMY M/AU

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E4            2      KUPPUSAMY M R/AU
E5            1      KUPPUSAMY MUSAVAN/AU
E6            2      KUPPUSAMY N/AU
E7            2      KUPPUSAMY NALLAGOUNDER/AU
E8          470      KUPPUSAMY P/AU
E9            1      KUPPUSAMY PARIANNAN/AU
E10           1      KUPPUSAMY PERIANNAM/AU
E11         269      KUPPUSAMY PERIANNAN/AU
E12           1      KUPPUSAMY R/AU

=> s e3
L7           40 "KUPPUSAMY M"/AU

=> e lahri s/au
E1            8      LAHRI RAJEEVA/AU
E2            1      LAHRI REJEEVA/AU
E3            5 --> LAHRI S/AU
E4            1      LAHRI S K/AU
E5            1      LAHRI V/AU
E6            1      LAHRI V L/AU
E7            2      LAHRICHI/AU
E8           13      LAHRICHI A/AU
E9            2      LAHRICHI ADIL/AU
E10           1      LAHRICHI ANDSIEST M G/AU
E11           4      LAHRICHI H/AU
E12           1      LAHRICHI H B/AU

=> s e3
L8            5 "LAHRI S"/AU

=> e khatri g s/au
E1           17      KHATRI G K/AU
E2           32      KHATRI G R/AU
E3           45 --> KHATRI G S/AU
E4            1      KHATRI GAJENDRA/AU
E5            1      KHATRI GAJENDRA K/AU
E6            1      KHATRI GAURAV/AU
E7            7      KHATRI GHAN SHYAM/AU
E8            2      KHATRI GOPAL KRISHAN/AU
E9            1      KHATRI GULSHAN R/AU
E10           3      KHATRI H/AU
E11           1      KHATRI H K/AU
E12          70      KHATRI H L/AU

=> s e3-e7
L9           55 ("KHATRI G S"/AU OR "KHATRI GAJENDRA"/AU OR "KHATRI GAJENDRA
                K"/AU OR "KHATRI GAURAV"/AU OR "KHATRI GHAN SHYAM"/AU)

=> s l7 or l8 or l9
L10         100 L7 OR L8 OR L9

=> d his

     (FILE 'HOME' ENTERED AT 11:36:50 ON 10 DEC 2004)

     FILE 'MEDLINE, EMBASE, BIOSIS, BIOTECHDS, SCISEARCH, HCAPLUS, NTIS,
     LIFESCI' ENTERED AT 11:37:16 ON 10 DEC 2004
L1         41164 S STREPTOKINASE?
L2           364 S "S. EQUISIMILIS"
L3           103 S L1 AND L2
L4       6828585 S CLON? OR EXPRESS? OR RECOMBINANT
L5            74 S L3 AND L4
L6            29 DUP REM L5 (45 DUPLICATES REMOVED)
                 E KUPPUSAMY M/AU
```

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L7            40 S E3
                 E LAHRI S/AU
L8             5 S E3
                 E KHATRI G S/AU
L9            55 S E3-E7
L10          100 S L7 OR L8 OR L9

=> s l5 and l10
L11           0 L5 AND L10

=> s l1 and l10
L12           0 L1 AND L10

=> d his

     (FILE 'HOME' ENTERED AT 11:36:50 ON 10 DEC 2004)

     FILE 'MEDLINE, EMBASE, BIOSIS, BIOTECHDS, SCISEARCH, HCAPLUS, NTIS,
     LIFESCI' ENTERED AT 11:37:16 ON 10 DEC 2004
L1         41164 S STREPTOKINASE?
L2           364 S "S. EQUISIMILIS"
L3           103 S L1 AND L2
L4       6828585 S CLON? OR EXPRESS? OR RECOMBINANT
L5            74 S L3 AND L4
L6            29 DUP REM L5 (45 DUPLICATES REMOVED)
                 E KUPPUSAMY M/AU
L7            40 S E3
                 E LAHRI S/AU
L8             5 S E3
                 E KHATRI G S/AU
L9            55 S E3-E7
L10          100 S L7 OR L8 OR L9
L11            0 S L5 AND L10
L12            0 S L1 AND L10
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	Issue Date	Pages	Document ID	Title
1	20041125	107	US 20040235011 A1	Production of multimeric proteins
2	20041007	174	US 20040197910 A1	Gene regulation in transgenic animals using a transposon-based vector
3	20040902	85	US 20040172667 A1	Administration of transposon-based vectors to reproductive organs
4	20031023	18	US 20030199810 A1	Methods and apparatuses for forming microprojection arrays
5	20030327	56	US 20030059921 A1	Novel clot-specific steptokinase proteins possessing altered plasminogen activation characteristics and a process for the preparation of said proteins
6	20010821	19	US RE37336 E	Method for providing hyaluronic acid
7	20010403	33	US 6210667 B1	Bacterial fibrin-dependent plasminogen activator
8	20000711	15	US 6087332 A	Streptokinase derivatives with high affinity for activated platelets and methods of their production and use in thrombolytic therapy
9	19981229	37	US 5854049 A	Plasmin-resistant streptokinase
10	19950718	72	US 5434073 A	Fibrinolytic and anti-thrombotic cleavable dimers
11	19940426	47	US 5306639 A	DNA encoding glucanase enzymes

	Issue Date	Pages	Document ID	Title
12	19940322	12	US 5296366 A	Method for the isolation and expression of a gene which codes for streptokinase, nucleotide sequence obtained, recombinant DNA and transformed microorgnaisms
13	19940301	47	US 5290916 A	Purified glucanase enzymes
14	19930817	10	US 5237050 A	Bacterial plasmin receptors as fibrinolytic agents
15	19930216	18	US 5187098 A	DNA encoding hybrid streptokinases with plasminogen fibrin binding domains
16	19911119	7	US 5066589 A	Streptokinase-coding recombinant vectors
17	19910514	19	US 5015577 A	DNA encoding hyaluronate synthase
18	19910430	18	US 5011686 A	Thrombus specific conjugates
19	19880816	7	US 4764469 A	Streptokinase-coding recombinant vectors

	L #	Hits	Search Text
1	L1	1	"5296366".pn.
2	L2	1	"5011686".pn.
3	L3	206	isoluble
4	L4	5961	inclusion adj bod$3
5	L5	0	l1 and l4
6	L6	0	l1 and l3
7	L7	0	streptokinsae$2
8	L8	4614	streptokinase$2
9	L9	0	"S. Equisimilis"
10	L10	16501	streptococcus
11	L11	71	l8 same l10
12	L12	685681	clon$3 or express$3 or recombinant
13	L13	19	l11 same l12
14	L14	499	KUPPUSAMY LAHRI KHATRI
15	L15	0	l13 and l14
16	L16	0	l11 and l14